Attorney's Docket No.: 12563-020001

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicant	: Wen-Tsan Chang et al.	Art Unit	: 1635
Serial No.	: 10/727,355	Examiner	: Dana H. Shin
Filed	: December 3, 2003	Conf. No.	: 5446
Title	: ANTIVIRUS RNA		

Mail Stop Amendment
Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

DECLARATION OF WEN-TSAN CHANG UNDER 37 C.F.R. § 1.131

I, Wen-Tsan Chang, hereby declare as follows:

1. I am a co-inventor of the subject matter described and claimed in the above-captioned application, which relates to an RNA molecule for inhibiting expression of a gene of a virus.

2. I have reviewed the office action, dated November 9, 2006, issued in this application. In this office action, claims 27-41 are rejected under 35 U.S.C. § 102(e) as being anticipated by Morrissey et al., US Patent Application Publication 2003/0206887 ("Morrissey"). Morrissey was filed on September 16, 2002.

3. On a date prior to September 16, 2002, Tsung-Lin Cheng, another co-inventor, sent a work order to MDBio Inc., Taipei, Taiwan ("MDBio"), requesting the synthesis of a pair of oligonucleotides, RNAi-HBVsAg-3-F and RNAi-HBVsAg-3-R, that contain SEQ ID NO:3 and its inverted repeat, respectively. A copy of the work order is attached hereto as "Exhibit A" with the order date blocked off. The oligonucleotides were received by us also on a date prior to September 16, 2002. A copy of the report from MDBio is attached hereto as "Exhibit B" with the received date also blocked off.

According to experimental records, the double-stranded DNA formed by RNAi-HBVsAg-3-F and RNAi-HBVsAg-3-R was ligated into a pSUPER vector on a date prior to September 16, 2002. A copy of the experimental records is attached hereto as "Exhibit C." The date entered by Tsung-Lin Cheng is blocked off. We proceeded to perform additional

experiments to reduce the invention to practice with due diligence and then have a provisional application prepared and filed on December 3, 2002 also with due diligence.

4. I hereby declare that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of the application or any patent issued thereon.

Respectfully submitted,

Date: February 15, 2007

Wen-Tsan Chang
Associate Professor
Department of Biochemistry and Molecular Biology, National Cheng Kung University Medical collage
1, University Road, Tainan 701, Taiwan, ROC

生工有限公司

Oligo 合成訂購單

單位：(請詳填院校系所及實驗室)
姓名：鄭琮霖　　電話：5528
日期：[redacted]　　頁次：共 1 頁之第 1 頁

為免混淆，A、T、C、請使用大寫字母，g 請使用小寫字母。
同位混合鹼基(equimolar mixing)請使用下列大寫字母代碼:

R: A,g	Y: C,T	M: A,C	K: g,T	S: g,C	W: A,T
H: A,T,C	B: g,T,C	V: g,A,C	D: g,A,T	N: A,T,G,C	

請自行給定 Oligo ID，並勾選適合您需要的項目。

Oligo ID： RNAi - HBVsAg - 3 - F　　☐. Thio-Oligo
☐. 40 nmole Scale　☑. 200 nmole Scale　　☐. PAGE Purification
Phosphorylation ☐. 5' ☐. 3'　　Biotin ☐. 5' ☐. 3'
AminoLink ☐. 5' ☐. 3'　　FITC ☐. 5' ☐. 3'

5' gATCC 05 CCggT 10 ATgTT 15 gCCCg 20
TTTgT 25 CTTCA 30 AgAgA 35 gACAA 40
ACggg 45 CAACA 50 TACCT 55 TTTTg 60 3'
gAAA

Oligo ID： RNAi - HBVsAg - 3 - R　　☐. Thio-Oligo
☐. 40 nmole Scale　☑. 200 nmole Scale　　☐. PAGE Purification
Phosphorylation ☐. 5' ☐. 3'　　Biotin ☐. 5' ☐. 3'
AminoLink ☐. 5' ☐. 3'　　FITC ☐. 5' ☐. 3'

5' AgCTT 05 TTCCA 10 AAAAg 15 gTATg 20
TTgCC 25 CgTTT 30 gTCTC 35 TCTTg 40
AAgAC 45 AAACg 50 ggCAA 55 CATAC 60 3'
Cggg

Oligo ID：　　☐. Thio-Oligo
☐. 40 nmole Scale　☐. 200 nmole Scale　　☐. PAGE Purification
Phosphorylation ☐. 5' ☐. 3'　　Biotin ☐. 5' ☐. 3'
AminoLink ☐. 5' ☐. 3'　　FITC ☐. 5' ☐. 3'

5' _____ 05 _____ 10 _____ 15 _____ 20
_____ 25 _____ 30 _____ 35 _____ 40
_____ 45 _____ 50 _____ 55 _____ 60 3'

生工有限公司　　免付費服務專線：080-072-222
電子郵件：mdbio@mdbio.neto.net　　傳真：(02)28238024

生工有限公司 MDBio Inc.

臺北市內湖區文德路 22 巷 9 弄 72 號 1 樓　免付費服務專線 080-072-222　電話：(02)27474876
傳真：(02)28238024　網站：www.mdbio.com.tw　電子郵件：mdbio@mdbio.com.tw

Oligo Primer 合成報告

Oligo ID: RNAi-H13USP9-5-F　　生產流水號：910706006

合成規模　☐40nmole　☑200nmole　☐1umole

純化方式　☐C18 脫鹽　☑PAGE 凝膠電泳

交貨總量：　10　OD_{260}　　每管含量：　5　OD_{260}

合成儀列印資料：

DNA SEQUENCE 1

NUMBER OF BASES:	64				
BASES USED:	A=17	C=15	G=15	T=17	X=0
DALTONS:	19634				
TIME:	18:48				
DATE:	[redacted]				

5' > GATCCCCGGTATGTTGCCCGTTTGTCTTCAAGAGAGACAAACGGGCAACA

TACCTTTTTGGAAA < 3'

- Oligo DNA 一般均以 OD_{260} 為計量單位，意指在光程 1 公分、吸收波長 260nm 時，吸光度為 1 的溶液 1 毫升內所含之 Oligo DNA 總量。據此定義，一 OD_{260} 單位相當於 33 微克(ug)的 Oligo DNA。
- 抽乾後之 Oligo DNA 為附著於管壁上之極輕薄膜，開啓管蓋時較易散失。開啓離心管前請先離心，並緩慢打開管蓋；溶解時請加足量水後蓋上管蓋，充份搖動 5 至 10 分鐘。
- 如需電泳分析 Oligo DNA，請採 1 倍 TBE Buffer 搭配含 7M 尿素之聚丙醯胺凝膠電泳，瓊脂糖凝膠電泳不適用於 Oligo DNA。另為避免加入電泳樣品時的擴散效應與二級結構影響電泳結果，電泳樣品請加飽和尿素處理。
- Oligo DNA 的兩端均為羥基，如需磷酸基，請另行處理或於合成時加選磷酸化處理。
- 溴乙基對單鏈 DNA 的染色效果受二級結構及長度影響甚鉅，等量的不同 Oligo DNA 染色後亮度可能差異極大。如電泳後溴乙基染色條帶亮度較弱，請將電泳凝膠置於紫外燈下之螢光板上觀察，一般可看到明亮的綠色螢光背景中含有清晰黑色條帶。如無前述操作所需設備，也可逕行以紫外光光度計檢測 OD_{260} 值。
- Oligo DNA 如欲長期儲存或暫不使用，請保存於-20℃。

生工有限公司 MDBio Inc.

臺北市內湖區文德路22巷9弄72號1樓　免付費服務專線080-072-222　電話:(02)27474876
傳真：(02)28238024　網站：www.mdbio.com.tw　電子郵件：mdbio@mdbio.com.tw

Oligo Primer 合成報告

Oligo ID: RNAi-ABUS19-3-R　　生產流水號：91070607

合成規模　☐40nmole　☑200nmole　☐1umole

純化方式　☐C18 脫鹽　☑PAGE 凝膠電泳

交貨總量：10 OD_{260}　　每管含量：5 OD_{260}

合成儀列印資料：

DNA SEQUENCE 1

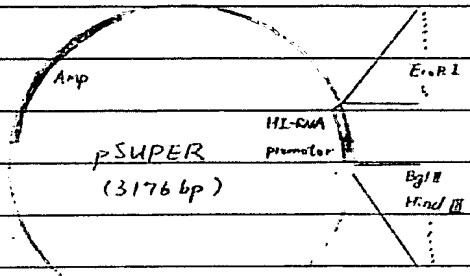
NUMBER OF BASES:	64
BASES USED:	A=17 C=15 G=15 T=17 X=0
DALTONS:	19634
TIME:	18:52
DATE:	[redacted]

5' > AGCTTTTCCAAAAAGGTATGTTGCCCGTTTGTCTCTCTTGAAGACAAACG

GGCAACATACCGGG < 3'

- Oligo DNA 一般均以 OD_{260} 為計量單位，意指在光程 1 公分、吸收波長 260nm 時，吸光度為 1 的溶液 1 毫升內所含之 Oligo DNA 總量。據此定義，一 OD_{260} 單位相當於 33 微克(ug)的 Oligo DNA。
- 抽乾後之 Oligo DNA 為附著於管壁上之極輕薄膜，開啓管蓋時較易散失。開啓離心管前請先離心，並緩慢打開管蓋；溶解時請加足量水後蓋上管蓋，充份搖動 5 至 10 分鐘。
- 如需電泳分析 Oligo DNA，請採 1 倍 TBE Buffer 搭配含 7M 尿素之聚丙醯胺凝膠電泳，瓊脂糖凝膠電泳不適用於 Oligo DNA。另為避免加入電泳樣品時的擴散效應與二級結構影響電泳結果，電泳樣品請加飽和尿素處理。
- Oligo DNA 的兩端均為羥基，如需磷酸基，請另行處理或於合成時加選磷酸化處理。
- 溴乙基對單鏈 DNA 的染色效果受二級結構及長度影響甚鉅，等量的不同 Oligo DNA 染色後亮度可能差異極大。如電泳後溴乙基染色條帶亮度較弱，請將電泳凝膠置於紫外燈下之螢光板上觀察，一般可看到明亮的綠色螢光背景中含有清晰黑色條帶。如無前述操作所需設備，也可逕行以紫外光光度計檢測 OD_{260} 值。
- Oligo DNA 如欲長期儲存或暫不使用，請保存於-20℃。

NO.
DATE [redacted]



RNAi - HBVsAg - 3 - F

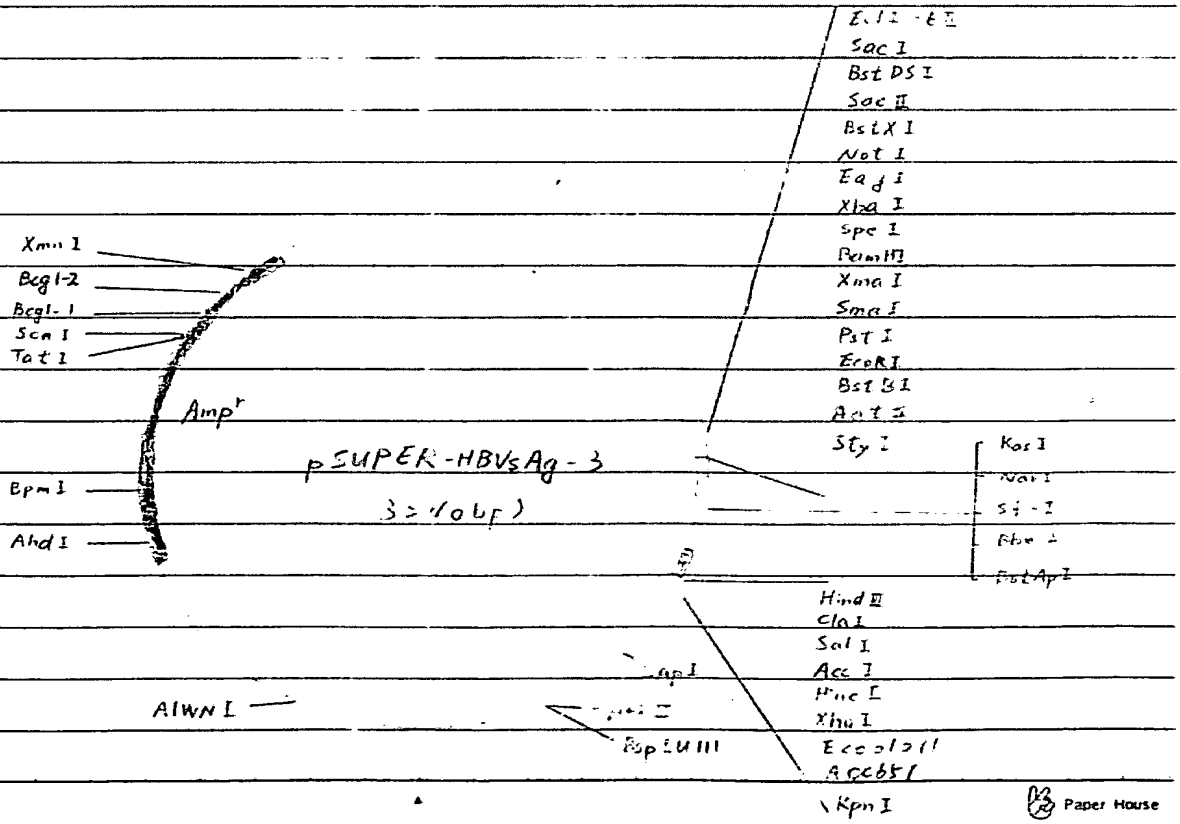
5' gATCC cggT ATgTT gcccg
TTTgT cTTCA AgAgA gAcAA
Acggg cAACA TACCT TTTTg
gAAA 3'

RNAi - HBVsAg - 3 - R

5' AgcTT TTCCA AAAAg gTATg
TTgcC cgTTT gTCTC TCTTg
AAgAC AAACg ggCAA cATAC
cgggg 3'

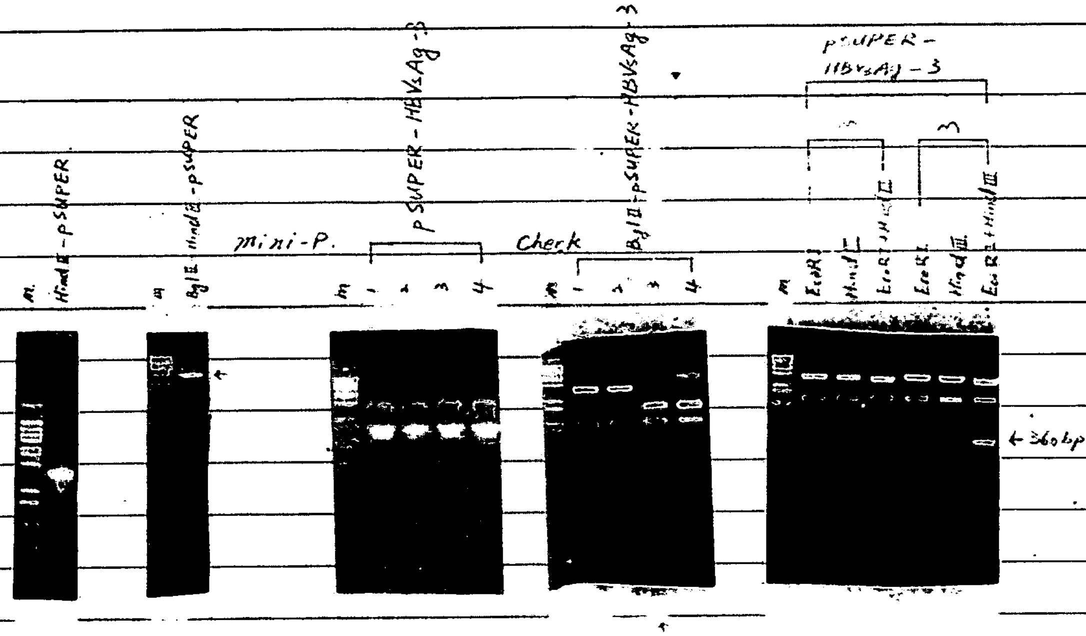
↓ Digest with Hind III
↓ Heat inactivation
↓ Phenol extraction
↓ Digest with Bgl II
↓ Gene Clean
Bgl II + Hind III - pSUPER (G.C)

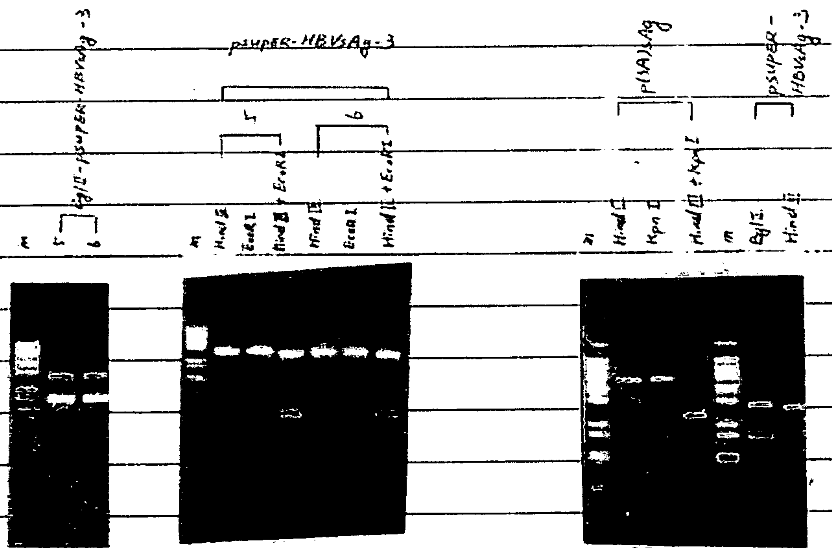
5' gATC [redacted] TCgA 5'



Paper House

NO.
DATE


m
HindIII - pSUPER
m
BglII HindIII - pSUPER
mini-P.
pSUPER - HBVsAg - 3
m 1 2 3 4
Check
BglII - pSUPER - HBVsAg - 3
m 1 2 3 4
pSUPER -
HBVsAg - 3
3
m
EcoRI
HindIII
EcoRI + HindIII
EcoRI
HindIII
EcoRI + HindIII
← 360bp


BglII - pSUPER - HBVsAg - 3
m 5 6
pSUPER - HBVsAg - 3
5
6
m
HindIII
EcoRI
HindIII + EcoRI
HindIII
EcoRI
HindIII + EcoRI
p(1A)sAg
m
HindIII
KpnI
HindIII + KpnI
pSUPER -
HBVsAg - 3
m
BglII
HindIII
DNA